

CLAIMS

1. An antibacterial protein which can be isolated from *S. salivarius* strain K12 on deposit at Deutsche Sammlung von Mikroorganismen Und Zellkulturen GmbH, Braunschweig, Germany, accession number DSM 13084, which has a molecular mass of approximately 2733 Da as determined by ion-spray mass spectrometry, and the N-terminal amino acid sequence of SEQ ID NO: 1, or an antibacterial fragment or variant thereof which variant has greater than 80% amino acid sequence homology with said protein.

2. An antibacterial protein having the amino acid sequence of SEQ ID NO: 3 or an antibacterial fragment or variant thereof, which variant has greater than 80% amino acid sequence homology with said protein.

3. An antibacterial protein having the amino acid sequence of SEQ ID NO: 3.

4. An antibacterial protein which has an amino acid sequence which differs from the sequence of SEQ ID NO 3 by the insertion, deletion or substitution of from one to three amino acids.

5. A protein as claimed in any one of claims 1-4 which is bacteriocidal.

6. A protein as claimed in claim 5 which is bacteriocidal with respect to *Streptococcus pyogenes*.

7. An antibacterial composition which includes a protein as claimed in any one of claims 1 to 6 or an organism which can express a protein as claimed in any one of claims 1 to 6.

8. A therapeutic formulation which comprises:

(i) a protein as claimed in any one of claims 1-6; or

(ii) an organism which can express a protein as claimed in any one of claims 1-6,

in combination with a diluent, carrier and/or excipient.

9. A therapeutic formulation according to claim 8 which comprises a protein as claimed in any one of claims 1-6 in combination with a diluent, carrier and/or excipient.
- 5 10. A therapeutic formulation as claimed in claim 8 which comprises an organism which can express a protein as claimed in any one of claims 1-6 in combination with a diluent, carrier and/or excipient.
- 10 11. A therapeutic formulation as claimed in any one of claims 8-10 which is an orally administrable medicament.
12. A medicament as claimed in claim 11 which is a syrup, mouthwash, gargle, toothpaste or mouth spray.
- 15 13. A medicament as claimed in claim 11 which is in a unit dosage form.
14. A medicament as claimed in claim 12 which is a lozenge or capsule containing a unit dose of an organism which can express a protein as claimed in any one of claims 1-6.
- 20 15. A therapeutic formulation as claimed in any one of claims 8-11 in which said protein or organism is included in a food or drink.
16. A formulation as claimed in claim 15 in which said food or drink is a dairy product based food or drink.
- 25 17. A formulation as claimed in claim 16 in which said protein or organism is included in milk powder, milk biscuits, milk, yoghurt or cheese.
- 30 18. A formulation as claimed in claim 16 in which said protein or organism is included in a flavoured milk.
19. A therapeutic formulation as claimed in any one of claims 8-10 in which said protein or organism is included in a confectionery.
- 35 20. A formulation as claimed in claim 19 in which said confectionery is a chewing gum.

21. A therapeutic formulation as claimed in any one of claims 9-20 which further comprises one or more secondary antibacterial agents.
- 5 22. A therapeutic formulation as claimed in claim 21 in which said secondary antibacterial agent(s) are selected from bacteriocin-like inhibitory substance(s) (BLIS).
- 10 23. A therapeutic formulation as claimed in claim 20 which includes one or both of Salivaricin A, an organism which can express Salivaricin A, the antibacterial protein as defined in claim 41 or an organism which can express the antibacterial protein as defined in claim 41.
- 15 24. A polynucleotide which encodes a protein as claimed in any one of claims 1-6.
- 20 25. A polynucleotide which comprises the coding sequence of SEQ ID NO: 2.
26. A polynucleotide as claimed in claim 24 which comprises a DNA sequence which encodes an antibacterial protein as claimed in claim 1 which is part of the genome of *S. salivarius* strain K12, on deposit at Deutsche Sammlung von Mikroorganismen Und Zellkulturen GmbH, Braunschweig, Germany, accession number DSM 13084.
- 25 27. An organism, in substantially pure form, which includes a polynucleotide as claimed in any one of claims 24-26 and is capable of expressing an antibacterial protein as claimed in any one of claims 1-6.
- 30 28. An organism as claimed in claim 27 in which said polynucleotide is heterologous.
29. An organism as claimed in claim 27 which is a *S. salivarius* organism.
- 35 30. A biologically pure culture of *S. salivarius* strain K12, on deposit at Deutsche Sammlung von Mikroorganismen Und Zellkulturen GmbH, Braunschweig, Germany, accession number DSM 13084.

31. A biologically pure culture of *S. salivarius* strain K30 on deposit at Deutsche Sammlung von Mikroorganismen Und Zellkulturen GmbH, Braunschweig, Germany, accession number DSM 13085.
- 5 32. A therapeutic formulation which includes *S. salivarius* strain K12 or *S. salivarius* strain K30 as identified in claim 30 or claim 31.
- 10 33. A method of treating an individual to at least inhibit growth of harmful streptococcal bacteria in the upper respiratory tract, comprising the step of administering an effective amount of a protein as claimed in any one of claims 1-6 orally to said individual.
- 15 34. A method as claimed in claim 33 in which said protein is administered orally to said individual as part of a therapeutic formulation as claimed in any one of claims 8 to 23 and 32.
- 20 35. A method as claimed in claim 33 wherein said inhibitory effect is caused by colonising at least part of the upper respiratory tract of an individual with a viable organism which expresses said protein.
- 25 36. A method as claimed in claim 35 wherein said organism is administered as part of a medicament, a food or drink or a confectionery.
- 30 37. A method as claimed in claim 35 or claim 36 wherein said organism is a *S. salivarius* strain selected from strains K12 and K30.
- 35 38. A method as claimed in any one of claims 33-37 which includes a preliminary step of pre-treating said individual to at least reduce the bacterial population present in the upper respiratory tract.
39. A method as claimed in claim 38 wherein said pre-treatment comprises the step of administering an antibiotic orally to said individual.
40. A method of treatment of a patient against infections of the upper respiratory tract caused by streptococcal organisms which comprises the steps of:

- (i) orally administering to said patient an amount of an antibiotic effective to reduce the numbers of streptococci present; and
- (ii) administering, to the resulting bacterially depopulated environment, *S. salivarius* organism(s) which produce BLIS to repopulate said environment.

41. An antibacterial protein which has the amino acid sequence of SEQ ID NO: 5.

42. A polynucleotide which includes the coding sequence of SEQ ID NO: 4.

43. A therapeutic formulation which comprises an antibacterial protein as claimed in claim 41, in combination with a diluent, carrier and/or excipient.

44. A therapeutic formulation which contains an antibacterial protein as claimed in any one of claims 1-6 and an antibacterial protein as claimed in claim 41.

45. A therapeutic formulation which contains at least one *S. salivarius* organism which expresses an antibacterial protein as claimed in any one of claims 1-6 and at least one other *S. salivarius* organism which expresses an antibacterial protein as claimed in claim 41.